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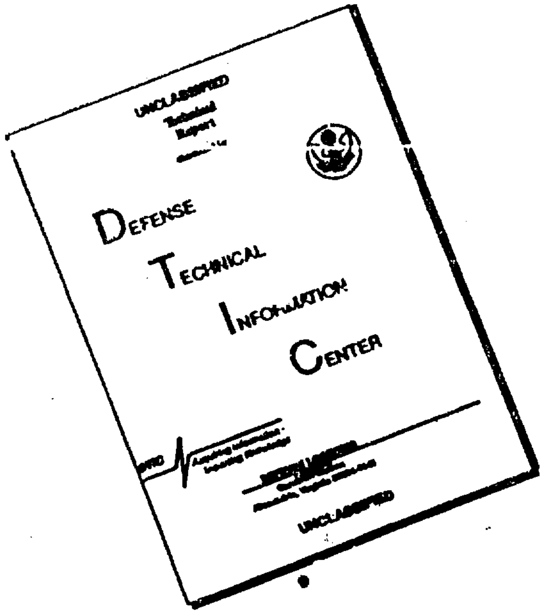

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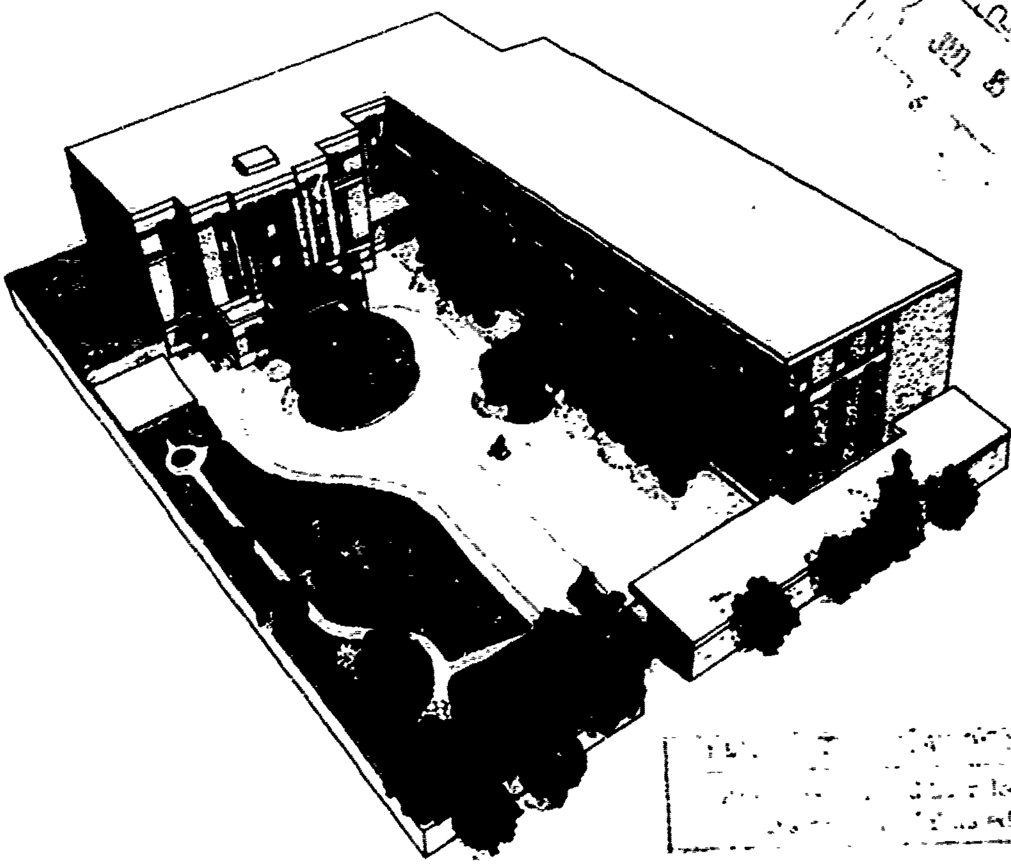
G-6-PD DEFICIENCY AND MALARIA IN BLACK AMERICANS IN VIETNAM



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13. ABSTRACT

ALTHOUGH THE SICKLE-CELL GENE IN NEGRO POPULATIONS HAS BEEN PROVEN TO CONFER RESISTANCE TO MALARIAL INFECTION, OTHER GENETIC MARKERS OCCURING PREDOMINANTLY IN NEGROES, INCLUDING GLUCOSE-6-PHOSPHATE DEHYDROGENASE (G-6-PD) DEFICIENCY, HAVE NOT BEEN SHOWN TO BE ASSOCIATED WITH A LOWER INCIDENCE OF MALARIA. HUMAN FEMALES HETEROZYGOUS FOR G-6-PD DEFICIENCY, WHICH IS A SEX-LINKED GENE, HAVE A MOSAICISM OF RED CELLS, AND THOSE CELLS DEFICIENT IN G-6-PD WERE SHOWN LESS LIKELY TO HARBOR P. FALCIPARUM PARASITES DURING AN INFECTION. THIS SUGGESTS THAT G-6-PD DEFICIENT MALES WHEN COMPARED WITH G-6-PD NORMAL MALES MIGHT HAVE MILDER CASES OF MALARIA, OR PERHAPS EVEN HAVE AN OVERALL LOWER INCIDENCE OF MALARIA INFECTION. THE PRESENT STUDY WAS UNDERTAKEN IN BLACK AMERICANS IN VIETNAM TO DETERMINE WHETHER G-6-PD DEFICIENCY HAS ANY INFLUENCE ON MALARIAL INCIDENCE OR THE CLINICAL SEVERITY OF MALARIAL INFECTIONS.

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G-6-PD Deficiency and Malaria in Black Americans in Vietnam

*LCDR Thomas Butler, MC, USNR**

Although the sickle-cell gene in Negro populations has been proven to confer resistance to malarial infection, other genetic markers occurring predominantly in Negroes, including glucose-6-phosphate dehydrogenase (G-6-PD) deficiency, have not been shown to be associated with a lower incidence of malaria.[2-5] Human females heterozygous for G-6-PD deficiency, which is a sex-linked gene, have a mosaicism of red cells, and those cells deficient in G-6-PD were shown less likely to harbor *P. falciparum* parasites during an infection.[6] This suggests that G-6-PD deficient males when compared with G-6-PD normal males might have milder cases of malaria, or perhaps even have an overall lower incidence of malaria infection. The present study was undertaken in Black Americans in Vietnam to determine whether G-6-PD deficiency has any influence on malarial incidence or the clinical severity of malarial infections.

Methods

Blood was obtained from 277 black subjects who were hospitalized between the months of February and July 1970 or visited the outpatient clinics of the NSA Station Hospital, Danang, the First Marines Medical Battalion, Danang, the USS Sanctuary, the Third Field Army Hospital, Saigon, the Sixth Convalescent Center, Cam Ranh Bay, and the 483rd USAF Hospital, Cam Ranh Bay. Most subjects were adhering by history to a malaria prophylactic regimen of weekly chloroquine (300 mg base) and primaquine (45 mg base). No specific selection was employed; all black patients on a given ward or in a given clinic on the day that the NAMRU-2 team visited the hospital who were willing to have blood taken were accepted. About 95 per cent of those asked consented to give blood. Hospital charts of malaria patients were read for results of malaria smears, the per cent of red cells parasitized, the lowest recorded hematocrit while in the hospital, renal function data, and fever duration if they had malaria. Subjects having a history of malaria in Vietnam were considered as malaria patients, and information from the previous hospitalization was recorded.

Blood was drawn into an oxalated Vacutainer and placed on ice. Within three hours of venepuncture, a modification of Motulsky's Brilliant Cresyl Blue dye test for G-6-PD activity was performed.[1] Decoloration times greater than 90 minutes were considered deficient. A hematocrit was performed whenever possible and was used as the lowest recorded hematocrit if none lower could be found in the subject's chart.

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A group of 28 white patients with malaria from the NSA Station Hospital was studied only for malaria species distribution.

Results

The overall G-6-PD deficiency rate was 38/277, or 13.7 per cent. Table I shows the distribution of diagnostic categories in all 277 subjects and the G-6-PD deficiency rate for each diagnosis. To determine whether G-6-PD deficiency may have influenced the rate of malaria infection in these patients, a comparison between the malaria and nonmalaria patients was made. Table II shows that only eight per cent of malaria patients had G-6-PD deficiency, while 17 per cent of the nonmalaria cases had this deficiency. The chi-square test applied to Table II revealed that, at a 95 per cent confidence level, the G-6-PD deficient group had a lower incidence of malaria (21 per cent) than did the G-6-PD normal group (39 per cent).

TABLE I

RESULTS OF G-6-PD DETERMINATIONS IN DIAGNOSTIC CATEGORIES FOR 277 BLACK SUBJECTS

	G-6-PD		
Diagnosis	Normal	Deficient	Total
P falciparum	76	7	83
P vivax	10	1	11
Mixed Malaria	5	0	5
Undetermined P. Species	2	0	2
Undiagnosed Fever	13	3	16
Virus Hepatitis	18	3	21
Diarrhea	14	3	17
Venereal Disease	11	1	12
Anemia	2	4	6
Seizure	2	1	3
Peptic Ulcer	2	1	3
Pneumonia	4	0	4
Typhoid Fever	2	0	2
Scrub Typhus	1	0	1
Cellulitis	2	0	2
Skin Abscess	2	0	2
Phlebitis	2	0	2
Diabetes	1	0	1
Varicella	0	1	1
Rheumatic Fever	1	0	1
Drug Overdose	4	2	6
Delirium Tremens	0	1	1
Headache	1	0	1
Other Medical Inpatients	16	4	20
Combat Injuries	18	2	20
Other Surgical Inpatients	17	4	21
Dermatology Clinic	3	0	3
Miscellaneous Outpatients	10	0	10
Total	239	38	277

The mean of the lowest recorded hematocrits in 19 of the 38 G-6-PD deficient subjects was 35.0 per cent, which was significantly lower than the 40.3 per cent for 131 of the 239 normal subjects (T = 4.05). Similarly, the malaria patients with G-6-PD deficiency had a significantly lower mean hematocrit (35.9 per cent) than did the malaria patients with normal G-6-PD activity (39.6 per cent) (T = 2.15).

Comparisons of the G-6-PD deficient malaria patients with the normal malaria patients in respect to clinical severity of disease, duration of fever, per cent of red cells parasitized, and BUN elevations did not reveal significant differences. None of the subjects developed renal failure or died while under observation.

The number of malaria patients among the black subjects was too small to determine whether the malaria-protective effect observed was greater for *P. falciparum* or *P. vivax* malaria. However, the proportion of pure *P. vivax* infections in the black malaria patients was compared with the group of 28 white malaria patients. The white patients had six *P. vivax* infections out of 28 total, or 21 per cent, while the blacks had 11 out of 102 cases, or a 11 per cent proportion of *P. vivax*, but this difference was not statistically significant.

Discussion

The lower incidence of malaria in the G-6-PD deficient subjects in this study suggests that a protection against malaria is associated with G-6-PD deficiency. However, possible selection biases must be considered. This study group was not randomly selected among all blacks in Vietnam, although no specific selection criteria other than hospitalization and availability were utilized. If G-6-PD deficiency influences the incidence of malaria or the other diseases listed in Table I, then selection biases will be present, particularly in the patients hospitalized on the medical wards. However, the overall incidence of 13.7 per cent G-6-PD deficiency in this study group, conforming with the black American average in the range of 10 per cent[6] to 13 per cent[2], indicates that no strong overall bias was present. Furthermore, comparisons of "medical" and "nonmedical" diseases did not reveal statistically significant differences in G-6-PD deficiency rates.

The other source of error in interpreting these results is the Brilliant Cresyl Blue dye screening test. It is a timed color change test, which depends on the amount of active enzyme present. Two types of error can occur: (1) False deficiencies, and (2) false normals. A G-6-PD normal individual with anemia will have fewer red cells, and hence less enzyme present in a given volume of whole blood, than a nonanemic individual will have. In this study, no compensation for hematocrit was made and, therefore, a few false deficiencies may be included. Conversely, because young red cells in G-6-PD deficient subjects contain more active G-6-PD enzyme than older cells, a deficient patient with compensated hemolysis may produce a normal test. Motulsky[7] believes that most errors are of the latter type. Because most of these study subjects were taking weekly primaquine as malaria prophylaxis, it would be expected that hemolysis caused by primaquine in the G-6-PD deficient subjects might result in some false normals. False normal results, if present in the malaria patients in this study, would give the illusion of malarial protection, by decreasing the true incidence of G-6-PD deficiency in the malaria patients.

TABLE II

CASE DISTRIBUTION SHOWING RELATIONSHIP BETWEEN MALARIA AND G-6-PD RESULTS

G-6-PD	Normal	Deficient	Total
Malaria	93	8	101
Non-malaria	146	30	176
Total	239	38	277

Chi-square = 4.51

Although the malaria patients with G-6-PD deficiency had clinical courses and severities of illness similar to the G-6-PD normal patients, the deficient patients as a group were more anemic. It appears unlikely that their more severe anemia was caused by a heavier parasitization, because the percentages of red cells parasitized in both groups were about the same. The more severe anemia in the G-6-PD deficient malaria patients was more likely caused by the oxidizing drugs, notably primaquine and aspirin, used to treat many of the patients. Other factors contributing to the hemolysis in deficient patients may have been the stress of a febrile disease[3] and the effect of hepatitis, which may occur in malaria.[4] Virus hepatitis may produce hemolysis in G-6-PD deficient patients by liberating metabolites toxic to the enzyme-deficient red cells,[8] and a similar mechanism might have occurred in these malaria patients.

In this study, it was not possible to correlate the malaria protective effect seen in G-6-PD deficient subjects with malarial species, because of the small number of subjects. The data in this study, suggesting that *P. vivax* occurred at a lower rate in black malaria patients than it did in white malaria patients, agree with other studies which demonstrate in Negroes a resistance to *P. vivax*.[5,10] However, the failure to show protection against *P. vivax* infections in G-6-PD deficient human volunteers[9] weighs against a specific *P. vivax* protection. The fact that most malaria patients in this study had *P. falciparum* infections, and that malaria occurred less frequently in G-6-PD deficient individuals, strongly suggests that malarial protection, if truly present, is being provided against *P. falciparum* malaria.

Summary

Of 277 black American male subjects in Vietnam tested for G-6-PD deficiency, 38 (13.7 per cent) were G-6-PD deficient. The incidence of malaria, predominantly *P. falciparum* malaria, was found to be significantly lower in the G-6-PD deficient blacks. Other than more severe anemia seen in the G-6-PD deficient malaria patients, no difference in the clinical severity of malaria was seen between the G-6-PD deficient and normal groups. These results are interpreted reservedly in light of possible selection biases and errors inherent in the G-6-PD determination.

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